



Sunesis-0021.ST25.txt
SEQUENCE LISTING

<110> ~~PatentIn~~ Finger, Marcus

<120> Constructs for Homogenously Processed Preparations of Beta Site
App-Cleaving Enzyme

<130> 2004345-0021

<140> 10/726,967

<141> 2003-12-02

<160> 110

<170> PatentIn version 3.2

<210> 1

<211> 501

<212> PRT

<213> Full length human BACE1 isform A

<400> 1

Met Ala Gln Ala Leu Pro Trp Leu Leu Leu Trp Met Gly Ala Gly Val
1 5 10 15

Leu Pro Ala His Gly Thr Gln His Gly Ile Arg Leu Pro Leu Arg Ser
20 25 30

Gly Leu Gly Gly Ala Pro Leu Gly Leu Arg Leu Pro Arg Glu Thr Asp
35 40 45

Glu Glu Pro Glu Glu Pro Gly Arg Arg Gly Ser Phe Val Glu Met Val
50 55 60

Asp Asn Leu Arg Gly Lys Ser Gly Gln Gly Tyr Tyr Val Glu Met Thr
65 70 75 80

Val Gly Ser Pro Pro Gln Thr Leu Asn Ile Leu Val Asp Thr Gly Ser
85 90 95

Ser Asn Phe Ala Val Gly Ala Ala Pro His Pro Phe Leu His Arg Tyr
100 105 110

Tyr Gln Arg Gln Leu Ser Ser Thr Tyr Arg Asp Leu Arg Lys Gly Val
115 120 125

Tyr Val Pro Tyr Thr Gln Gly Lys Trp Glu Gly Glu Leu Gly Thr Asp
130 135 140

Leu Val Ser Ile Pro His Gly Pro Asn Val Thr Val Arg Ala Asn Ile
145 150 155 160

Sunesis-0021.ST25.txt

Ala Ala Ile Thr Glu Ser Asp Lys Phe Phe Ile Asn Gly Ser Asn Trp
165 170

Glu Gly Ile Leu Gly Leu Ala Tyr Ala Glu Ile Ala Arg Pro Asp Asp
180 185 190

Ser Leu Glu Pro Phe Phe Asp Ser Leu Val Lys Gln Thr His Val Pro
195 200 205

Asn Leu Phe Ser Leu Gln Leu Cys Gly Ala Gly Phe Pro Leu Asn Gln
210 215 220

Ser Glu Val Leu Ala Ser Val Gly Gly Ser Met Ile Ile Gly Gly Ile
225 230 235 240

Asp His Ser Leu Tyr Thr Gly Ser Leu Trp Tyr Thr Pro Ile Arg Arg
245 250 255

Glu Trp Tyr Tyr Glu Val Ile Ile Val Arg Val Glu Ile Asn Gly Gln
260 265 270

Asp Leu Lys Met Asp Cys Lys Glu Tyr Asn Tyr Asp Lys Ser Ile Val
275 280 285

Asp Ser Gly Thr Thr Asn Leu Arg Leu Pro Lys Lys Val Phe Glu Ala
290 295 300

Ala Val Lys Ser Ile Lys Ala Ala Ser Ser Thr Glu Lys Phe Pro Asp
305 310 315 320

Gly Phe Trp Leu Gly Glu Gln Leu Val Cys Trp Gln Ala Gly Thr Thr
325 330 335

Pro Trp Asn Ile Phe Pro Val Ile Ser Leu Tyr Leu Met Gly Glu Val
340 345 350

Thr Asn Gln Ser Phe Arg Ile Thr Ile Leu Pro Gln Gln Tyr Leu Arg
355 360 365

Pro Val Glu Asp Val Ala Thr Ser Gln Asp Asp Cys Tyr Lys Phe Ala
370 375 380

Ile Ser Gln Ser Ser Thr Gly Thr Val Met Gly Ala Val Ile Met Glu
385 390 395 400

Gly Phe Tyr Val Val Phe Asp Arg Ala Arg Lys Arg Ile Gly Phe Ala
405 410 415

Sunesis-0021.ST25.txt

Val Ser Ala Cys His Val His Asp Glu Phe Arg Thr Ala Ala Val Glu
420 425 430

Gly Pro Phe Val Thr Leu Asp Met Glu Asp Cys Gly Tyr Asn Ile Pro
435 440 445

Gln Thr Asp Glu Ser Thr Leu Met Thr Ile Ala Tyr Val Met Ala Ala
450 455 460

Ile Cys Ala Leu Phe Met Leu Pro Leu Cys Leu Met Val Cys Gln Trp
465 470 475 480

Arg Cys Leu Arg Cys Leu Arg Gln Gln His Asp Asp Phe Ala Asp Asp
485 490 495

Ile Ser Leu Leu Lys
500

<210> 2
<211> 10
<212> PRT
<213> Beta secretase cleavage site of Swedish mutant of human amyloid precursor protein (APP)

<400> 2

Ser Glu Val Asn Leu Asp Ala Glu Phe Arg
1 5 10

<210> 3
<211> 16
<212> PRT
<213> Residues 22-37 of human BACE1 preprosequence

<400> 3

Thr Gln His Gly Ile Arg Leu Pro Leu Arg Ser Gly Leu Gly Gly Ala
1 5 10 15

<210> 4
<211> 6
<212> PRT
<213> Preferred thrombin cleavage site

<400> 4

Leu Val Pro Arg Gly Ser
1 5

<210> 5
<211> 7
<212> PRT
<213> TEV protease cleavage site

<220>
<221> misc_feature
<222> (7)..(7)
<223> Xaa can be any naturally occurring amino acid

<400> 5

Glu Asn Leu Tyr Phe Asn Xaa
1 5

<210> 6
<211> 7
<212> PRT
<213> Preferred TEV protease cleavage site

<400> 6

Glu Asn Leu Tyr Phe Asn Gly
1 5

<210> 7
<211> 7
<212> PRT
<213> Preferred TEV protease cleavage site

<400> 7

Glu Asn Leu Tyr Phe Asn Ala
1 5

<210> 8
<211> 5
<212> PRT
<213> Genenase I cleavage site

<220>
<221> misc_feature
<222> (2)..(2)
<223> Xaa can be any naturally occurring amino acid

<400> 8

Ala Xaa His Tyr Ala
1 5

<210> 9
<211> 5
<212> PRT
<213> Genenase I cleavage site

<220>
<221> misc_feature
<222> (2)..(2)
<223> Xaa can be any naturally occurring amino acid

<400> 9

Ala Xaa His Phe Ala
1 5

<210> 10

<211> 5

<212> PRT

<213> Genenase I cleavage site

<220>

<221> misc_feature

<222> (2)..(2)

<223> Xaa can be any naturally occurring amino acid

<400> 10

Ala Xaa His Leu Ala
1 5

<210> 11

<211> 5

<212> PRT

<213> Genenase I cleavage site

<220>

<221> misc_feature

<222> (2)..(2)

<223> Xaa can be any naturally occurring amino acid

<400> 11

Phe Xaa His Tyr Ala
1 5

<210> 12

<211> 5

<212> PRT

<213> Genenase I cleavage site

<220>

<221> misc_feature

<222> (2)..(2)

<223> Xaa can be any naturally occurring amino acid

<400> 12

Phe Xaa His Phe Ala
1 5

<210> 13

<211> 5

<212> PRT

<213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 13

Phe Xaa His Leu Ala
 1 5

<210> 14
 <211> 5
 <212> PRT
 <213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 14

Leu Xaa His Tyr Ala
 1 5

<210> 15
 <211> 5
 <212> PRT
 <213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 15

Leu Xaa His Phe Ala
 1 5

<210> 16
 <211> 5
 <212> PRT
 <213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 16

Leu Xaa His Leu Ala
 1 5

<210> 17

<211> 5
 <212> PRT
 <213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 17

Tyr Xaa His Tyr Ala
 1 5

<210> 18
 <211> 5
 <212> PRT
 <213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 18

Tyr Xaa His Phe Ala
 1 5

<210> 19
 <211> 5
 <212> PRT
 <213> Genenase I cleavage site

<220>
 <221> misc_feature
 <222> (2)..(2)
 <223> Xaa can be any naturally occurring amino acid
 <400> 19

Tyr Xaa His Leu Ala
 1 5

<210> 20
 <211> 7
 <212> PRT
 <213> Preferred genenase I cleavage site
 <400> 20

Pro Gly Ala Ala His Tyr Ala
 1 5

<210> 21
 <211> 5

<212> PRT
<213> preferred enterokinase cleavage site

<400> 21

Asp Asp Asp Asp Lys
1 5

<210> 22
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site

<400> 22

Asp Glu Asp Asp Lys
1 5

<210> 23
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site

<400> 23

Asp Asp Glu Asp Lys
1 5

<210> 24
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site

<400> 24

Asp Asp Asp Glu Lys
1 5

<210> 25
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site

<400> 25

Glu Asp Asp Asp Lys
1 5

<210> 26
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site

<400> 26

Asp Glu Glu Asp Lys
1 5

<210> 27
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 27

Asp Glu Asp Glu Lys
1 5

<210> 28
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 28

Asp Asp Glu Glu Lys
1 5

<210> 29
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 29

Glu Asp Asp Glu Lys
1 5

<210> 30
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 30

Glu Glu Asp Asp Lys
1 5

<210> 31
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 31

Glu Asp Glu Asp Lys
1 5

<210> 32
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 32

Glu Glu Glu Asp Lys
1 5

<210> 33
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 33

Glu Glu Asp Glu Lys
1 5

<210> 34
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 34

Glu Asp Glu Glu Lys
1 5

<210> 35
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 35

Asp Glu Glu Glu Lys
1 5

<210> 36
<211> 5
<212> PRT
<213> preferred enterokinase cleavage site
<400> 36

Glu Glu Glu Glu Lys
1 5

<210> 37
<211> 6
<212> PRT
<213> preferred granzyme B cleavage site

<220>
<221> misc_feature
<222> (3)..(3)
<223> xaa can be any naturally occurring amino acid

<220>
<221> misc_feature
<222> (5)..(5)
<223> xaa can be any naturally occurring amino acid

<400> 37

Ile Glu Xaa Asp Xaa Gly
1 5

<210> 38

<211> 6

<212> PRT

<213> preferred granzyme B cleavage site

<220>

<221> misc_feature

<222> (3)..(3)

<223> Xaa can be any naturally occurring amino acid

<220>

<221> misc_feature

<222> (5)..(5)

<223> Xaa can be any naturally occurring amino acid

<400> 38

Ile Glu Xaa Asp Xaa Ala
1 5

<210> 39

<211> 6

<212> PRT

<213> preferred granzyme B cleavage site

<220>

<221> misc_feature

<222> (3)..(3)

<223> Xaa can be any naturally occurring amino acid

<220>

<221> misc_feature

<222> (5)..(5)

<223> Xaa can be any naturally occurring amino acid

<400> 39

Ile Met Xaa Asp Xaa Gly
1 5

<210> 40

<211> 6

<212> PRT

<213> preferred granzyme B cleavage site

<220>

<221> misc_feature

<222> (3)..(3)

<223> Xaa can be any naturally occurring amino acid

<220>

<221> misc_feature
 <222> (5)..(5)
 <223> Xaa can be any naturally occurring amino acid
 <400> 40

Ile Met Xaa Asp Xaa Ala
 1 5

<210> 41
 <211> 6
 <212> PRT
 <213> preferred granzyme B cleavage site

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> Xaa can be any naturally occurring amino acid

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> Xaa can be any naturally occurring amino acid
 <400> 41

Ile Gln Xaa Asp Xaa Gly
 1 5

<210> 42
 <211> 6
 <212> PRT
 <213> preferred granzyme B cleavage site

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> Xaa can be any naturally occurring amino acid

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> Xaa can be any naturally occurring amino acid
 <400> 42

Ile Gln Xaa Asp Xaa Ala
 1 5

<210> 43
 <211> 6
 <212> PRT
 <213> preferred granzyme B cleavage site

<220>
 <221> misc_feature
 <222> (3)..(3)

<223> Xaa can be any naturally occurring amino acid

<220>

<221> misc_feature

<222> (5)..(5)

<223> Xaa can be any naturally occurring amino acid

<400> 43

Val Glu Xaa Asp Xaa Gly
1 5

<210> 44

<211> 6

<212> PRT

<213> preferred granzyme B cleavage site

<220>

<221> misc_feature

<222> (3)..(3)

<223> Xaa can be any naturally occurring amino acid

<220>

<221> misc_feature

<222> (5)..(5)

<223> Xaa can be any naturally occurring amino acid

<400> 44

Val Glu Xaa Asp Xaa Ala
1 5

<210> 45

<211> 6

<212> PRT

<213> preferred granzyme B cleavage site

<220>

<221> misc_feature

<222> (3)..(3)

<223> Xaa can be any naturally occurring amino acid

<220>

<221> misc_feature

<222> (5)..(5)

<223> Xaa can be any naturally occurring amino acid

<400> 45

Val Met Xaa Asp Xaa Gly
1 5

<210> 46

<211> 6

<212> PRT

<213> preferred granzyme B cleavage site

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> Xaa can be any naturally occurring amino acid

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> Xaa can be any naturally occurring amino acid

<400> 46

Val Met Xaa Asp Xaa Ala
 1 5

<210> 47
 <211> 6
 <212> PRT
 <213> preferred granzyme B cleavage site

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> Xaa can be any naturally occurring amino acid

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> Xaa can be any naturally occurring amino acid

<400> 47

Val Gln Xaa Asp Xaa Gly
 1 5

<210> 48
 <211> 6
 <212> PRT
 <213> preferred granzyme B cleavage site

<220>
 <221> misc_feature
 <222> (3)..(3)
 <223> Xaa can be any naturally occurring amino acid

<220>
 <221> misc_feature
 <222> (5)..(5)
 <223> Xaa can be any naturally occurring amino acid

<400> 48

Val Gln Xaa Asp Xaa Ala
 1 5

<210> 49
 <211> 5
 <212> PRT

<213> Preferred turnip mosaic virus protease N1a cleavage site

<400> 49

Val Arg His Gln Ser
1 5

<210> 50

<211> 4

<212> PRT

<213> Preferred Fxator Xa cleavage site

<400> 50

Ile Asp Gly Arg
1

<210> 51

<211> 4

<212> PRT

<213> Preferred Fxator Xa cleavage site

<400> 51

Ile Glu Gly Arg
1

<210> 52

<211> 28

<212> PRT

<213> Residues 74-101 of human BACE1 preprosequence

<400> 52

Gly Tyr Tyr Val Glu Met Thr Val Gly Ser Pro Pro Gln Thr Leu Asn
1 5 10 15

Ile Leu Val Asp Thr Gly Ser Ser Asn Phe Ala Val
20 25

<210> 53

<211> 28

<212> PRT

<213> Residues 118-145 of human BACE1 preprosequence

<400> 53

Ser Ser Thr Tyr Arg Asp Leu Arg Lys Gly Val Tyr Val Pro Tyr Thr
1 5 10 15

Gln Gly Lys Trp Glu Gly Glu Leu Gly Thr Asp Leu
20 25

<210> 54

<211> 21

<212> PRT

Sunesis-0021.ST25.txt

<213> Residues 165-185 of Human BACE1 Preprosequence isoforms A and B

<400> 54

Glu Ser Asp Lys Phe Phe Ile Asn Gly Ser Asn Trp Glu Gly Ile Leu
1 5 10 15

Gly Leu Ala Tyr Ala
20

<210> 55

<211> 19

<212> PRT

<213> Residues 280-298 of human BACE1 preprosequence isoform A

<400> 55

Glu Tyr Asn Tyr Asp Lys Ser Ile Val Asp Ser Gly Thr Thr Asn Leu
1 5 10 15

Arg Leu Pro

<210> 56

<211> 7

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 56

Glu Leu Asn Leu Glu Thr Asp
1 5

<210> 57

<211> 7

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 57

Glu Ile Asn Leu Glu Thr Asp
1 5

<210> 58

<211> 7

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 58

Glu Ile Asn Phe Glu Thr Asp
1 5

<210> 59

<211> 7

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 59

Glu Val Asn Leu Asp Ala Glu
1 5

<210> 60

<211> 8

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 60

Glu Ile Asn Phe Ser Phe Val Glu
1 5

<210> 61

<211> 8

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 61

Glu Ile Asn Phe Gln Phe Val Asp
1 5

<210> 62

<211> 8

<212> PRT

<213> Engineered BACE1 autoproteolysis site

<400> 62

Glu Ile Asn Phe Ser Ala Ser Phe
1 5

<210> 63

<211> 6

<212> PRT

<213> Residues 53-58 of human preproBACE1

<400> 63

Glu Pro Gly Arg Arg Gly
1 5

<210> 64

<211> 24

<212> DNA

<213> Primer used to obtain bases 126-374 of human BACE1, and bases 126-1362 of human BACE1

<400> 64

gctgccccgg gagaccgacg aaga

24

<210> 65

Sunesis-0021.ST25.txt

<211> 24
 <212> DNA
 <213> Primer used to obtain nucleotides 126-374 of human BACE1

<400> 65
 cggagggtccc ggtatgtgct ggac 24

<210> 66
 <211> 24
 <212> DNA
 <213> Primer used to obtain nucleotides 339-770 of human BACE1

<400> 66
 ccagaggcag ctgtccagca cata 24

<210> 67
 <211> 24
 <212> DNA
 <213> Primer used to obtain nucleotides 339-770 of human BACE1

<400> 67
 tcccgccgga tgggtgtata ccag 24

<210> 68
 <211> 27
 <212> DNA
 <213> Primer used to obtain nucleotides 735-1551 of human BACE1

<400> 68
 gtacacaggc agtctctggt atacacc 27

<210> 69
 <211> 29
 <212> DNA
 <213> Primer used to obtain nucleotides 735-1551 of human BACE1

<400> 69
 gtgtggtcca ggggaatctc tatcttctg 29

<210> 70
 <211> 40
 <212> DNA
 <213> Primer containing an XhoI restriction site used to obtain nucleotides 126-1551 of human BACE1

<400> 70
 gtcacgtct cgagtcactt cagcagggag atgtcatcag 40

<210> 71
 <211> 60
 <212> DNA
 <213> Forward primer used to extend nucleotides 126-1362 of human BACE1 in 5' direction

<400> 71
 cggtgcccc tgcgcagcgg cctggggggc gccccctgg ggctgcggct gccccgggag 60

Sunesis-0021.ST25.txt

<210> 72
 <211> 60
 <212> DNA
 <213> Forward primer used to extend nucleotides 126-1362 of human BACE1 in 5' direction

<400> 72
 atgggcgcgg gagtgctgcc tgccacggc acccagcacg gcatccggct gcccctgcgc 60

<210> 73
 <211> 57
 <212> DNA
 <213> Forward primer containing an EcoRI restriction site used to extend nucleotides 126-1362 of human BACE1 in 5' direction

<400> 73
 ccggaattca tggccaagc cctgccctgg ctctgctgt ggatgggcgc gggagtg 57

<210> 74
 <211> 33
 <212> DNA
 <213> Forward primer containing an NdeI restriction site used to obtain bases 1-1362 of human BACE1

<400> 74
 cgccatatgg cgggagtgt gcctgcccac ggc 33

<210> 75
 <211> 36
 <212> DNA
 <213> Reverse primer containing an EcoRI restriction site used to obtain bases 1-1362 of human BACE1

<400> 75
 ccggaattct caggttgact catctgtctg tggaat 36

<210> 76
 <211> 439
 <212> PRT
 <213> Soluble human pro BACE1 with engineered thrombin cleavage site starting at position 25

<400> 76
 Thr Gln His Gly Ile Arg Leu Pro Leu Arg Ser Gly Leu Gly Gly Ala
 1 5 10 15

Pro Leu Gly Leu Arg Leu Pro Arg Leu Val Pro Arg Gly Ser Glu Thr
 20 25 30

Asp Glu Glu Pro Glu Glu Pro Gly Arg Arg Gly Ser Phe Val Glu Met
 35 40 45

Val Asp Asn Leu Arg Gly Lys Ser Gly Gln Gly Tyr Tyr Val Glu Met
 50 55 60

Sunesis-0021.ST25.txt

Thr Val Gly Ser Pro Pro Gln Thr Leu Asn Ile Leu Val Asp Thr Gly
65 70 75 80

Ser Ser Asn Phe Ala Val Gly Ala Ala Pro His Pro Phe Leu His Arg
85 90 95

Tyr Tyr Gln Arg Gln Leu Ser Ser Thr Tyr Arg Asp Leu Arg Lys Gly
100 105 110

Val Tyr Val Pro Tyr Thr Gln Gly Lys Trp Glu Gly Glu Leu Gly Thr
115 120 125

Asp Leu Val Ser Ile Pro His Gly Pro Asn Val Thr Val Arg Ala Asn
130 135 140

Ile Ala Ala Ile Thr Glu Ser Asp Lys Phe Phe Ile Asn Gly Ser Asn
145 150 155 160

Trp Glu Gly Ile Leu Gly Leu Ala Tyr Ala Glu Ile Ala Arg Pro Asp
165 170 175

Asp Ser Leu Glu Pro Phe Phe Asp Ser Leu Val Lys Gln Thr His Val
180 185 190

Pro Asn Leu Phe Ser Leu Gln Leu Cys Gly Ala Gly Phe Pro Leu Asn
195 200 205

Gln Ser Glu Val Leu Ala Ser Val Gly Gly Ser Met Ile Ile Gly Gly
210 215 220

Ile Asp His Ser Leu Tyr Thr Gly Ser Leu Trp Tyr Thr Pro Ile Arg
225 230 235 240

Arg Glu Trp Tyr Tyr Glu Val Ile Ile Val Arg Val Glu Ile Asn Gly
245 250 255

Gln Asp Leu Lys Met Asp Cys Lys Glu Tyr Asn Tyr Asp Lys Ser Ile
260 265 270

Val Asp Ser Gly Thr Thr Asn Leu Arg Leu Pro Lys Lys Val Phe Glu
275 280 285

Ala Ala Val Lys Ser Ile Lys Ala Ala Ser Ser Thr Glu Lys Phe Pro
290 295 300

Asp Gly Phe Trp Leu Gly Glu Gln Leu Val Cys Trp Gln Ala Gly Thr
Page 20

305 310 315 320

Thr Pro Trp Asn Ile Phe Pro Val Ile Ser Leu Tyr Leu Met Gly Glu
325 330 335

Val Thr Asn Gln Ser Phe Arg Ile Thr Ile Leu Pro Gln Gln Tyr Leu
340 345 350

Arg Pro Val Glu Asp Val Ala Thr Ser Gln Asp Asp Cys Tyr Lys Phe
355 360 365

Ala Ile Ser Gln Ser Ser Thr Gly Thr Val Met Gly Ala Val Ile Met
370 375 380

Glu Gly Phe Tyr Val Val Phe Asp Arg Ala Arg Lys Arg Ile Gly Phe
385 390 395 400

Ala Val Ser Ala Cys His Val His Asp Glu Phe Arg Thr Ala Ala Val
405 410 415

Glu Gly Pro Phe Val Thr Leu Asp Met Glu Asp Cys Gly Tyr Asn Ile
420 425 430

Pro Gln Thr Asp Glu Ser Thr
435

<210> 77

<211> 48

<212> DNA

<213> Oligonucleotide for insertion of thrombin cleavage site into soluble human proBACE1

<400> 77

ctcttcgctg gtctcagaac cgcgcggaac cagacgtggc agacgcag

48

<210> 78

<211> 433

<212> PRT

<213> Soluble human proBACE1 containing an engineered RLPL site starting at position 21

<400> 78

Thr Gln His Gly Ile Arg Leu Pro Leu Arg Ser Gly Leu Gly Gly Ala
1 5 10 15

Pro Leu Gly Leu Arg Leu Pro Leu Glu Thr Asp Glu Glu Pro Glu Glu
20 25 30

Pro Gly Arg Arg Gly Ser Phe Val Glu Met Val Asp Asn Leu Arg Gly
35 40 45

Sunesis-0021.ST25.txt

Lys Ser Gly Gln Gly Tyr Tyr Val Glu Met Thr Val Gly Ser Pro Pro
 50 55 60
 Gln Thr Leu Asn Ile Leu Val Asp Thr Gly Ser Ser Asn Phe Ala Val
 65 70 75 80
 Gly Ala Ala Pro His Pro Phe Leu His Arg Tyr Tyr Gln Arg Gln Leu
 85 90 95
 Ser Ser Thr Tyr Arg Asp Leu Arg Lys Gly Val Tyr Val Pro Tyr Thr
 100 105 110
 Gln Gly Lys Trp Glu Gly Glu Leu Gly Thr Asp Leu Val Ser Ile Pro
 115 120 125
 His Gly Pro Asn Val Thr Val Arg Ala Asn Ile Ala Ala Ile Thr Glu
 130 135 140
 Ser Asp Lys Phe Phe Ile Asn Gly Ser Asn Trp Glu Gly Ile Leu Gly
 145 150 155 160
 Leu Ala Tyr Ala Glu Ile Ala Arg Pro Asp Asp Ser Leu Glu Pro Phe
 165 170 175
 Phe Asp Ser Leu Val Lys Gln Thr His Val Pro Asn Leu Phe Ser Leu
 180 185 190
 Gln Leu Cys Gly Ala Gly Phe Pro Leu Asn Gln Ser Glu Val Leu Ala
 195 200 205
 Ser Val Gly Gly Ser Met Ile Ile Gly Gly Ile Asp His Ser Leu Tyr
 210 215 220
 Thr Gly Ser Leu Trp Tyr Thr Pro Ile Arg Arg Glu Trp Tyr Tyr Glu
 225 230 235 240
 Val Ile Ile Val Arg Val Glu Ile Asn Gly Gln Asp Leu Lys Met Asp
 245 250 255
 Cys Lys Glu Tyr Asn Tyr Asp Lys Ser Ile Val Asp Ser Gly Thr Thr
 260 265 270
 Asn Leu Arg Leu Pro Lys Lys Val Phe Glu Ala Ala Val Lys Ser Ile
 275 280 285
 Lys Ala Ala Ser Ser Thr Glu Lys Phe Pro Asp Gly Phe Trp Leu Gly
 Page 22

290

295

Glu Gln Leu Val Cys Trp Gln Ala Gly Thr Thr Pro Trp Asn Ile Phe
305 310 315 320
Pro Val Ile Ser Leu Tyr Leu Met Gly Glu Val Thr Asn Gln Ser Phe
325 330 335
Arg Ile Thr Ile Leu Pro Gln Gln Tyr Leu Arg Pro Val Glu Asp Val
340 345 350
Ala Thr Ser Gln Asp Asp Cys Tyr Lys Phe Ala Ile Ser Gln Ser Ser
355 360 365
Thr Gly Thr Val Met Gly Ala Val Ile Met Glu Gly Phe Tyr Val Val
370 375 380
Phe Asp Arg Ala Arg Lys Arg Ile Gly Phe Ala Val Ser Ala Cys His
385 390 395 400
Val His Asp Glu Phe Arg Thr Ala Ala Val Glu Gly Pro Phe Val Thr
405 410 415
Leu Asp Met Glu Asp Cys Gly Tyr Asn Ile Pro Gln Thr Asp Glu Ser
420 425 430

Thr

<210> 79
<211> 7
<212> PRT
<213> RLPL site

<400> 79

Arg Leu Pro Leu Glu Thr Asp
1 5

<210> 80
<211> 42
<212> DNA
<213> Oligonucleotide to introduce RLPL site into soluble human proBACE1

<400> 80
ctcttcgtcg gtctccagtg gcagacgcag acccagtgga gc 42

<210> 81
<211> 433
<212> PRT
<213> Soluble human proBACE1

<400> 81

Thr Gln His Gly Ile Arg Leu Pro Leu Arg Ser Gly Leu Gly Gly Ala
1 5 10 15

Pro Leu Gly Leu Glu Leu Asn Leu Glu Thr Asp Glu Glu Pro Glu Glu
20 25 30

Pro Gly Arg Arg Gly Ser Phe Val Glu Met Val Asp Asn Leu Arg Gly
35 40 45

Lys Ser Gly Gln Gly Tyr Tyr Val Glu Met Thr Val Gly Ser Pro Pro
50 55 60

Gln Thr Leu Asn Ile Leu Val Asp Thr Gly Ser Ser Asn Phe Ala Val
65 70 75 80

Gly Ala Ala Pro His Pro Phe Leu His Arg Tyr Tyr Gln Arg Gln Leu
85 90 95

Ser Ser Thr Tyr Arg Asp Leu Arg Lys Gly Val Tyr Val Pro Tyr Thr
100 105 110

Gln Gly Lys Trp Glu Gly Glu Leu Gly Thr Asp Leu Val Ser Ile Pro
115 120 125

His Gly Pro Asn Val Thr Val Arg Ala Asn Ile Ala Ala Ile Thr Glu
130 135 140

Ser Asp Lys Phe Phe Ile Asn Gly Ser Asn Trp Glu Gly Ile Leu Gly
145 150 155 160

Leu Ala Tyr Ala Glu Ile Ala Arg Pro Asp Asp Ser Leu Glu Pro Phe
165 170 175

Phe Asp Ser Leu Val Lys Gln Thr His Val Pro Asn Leu Phe Ser Leu
180 185 190

Gln Leu Cys Gly Ala Gly Phe Pro Leu Asn Gln Ser Glu Val Leu Ala
195 200 205

Ser Val Gly Gly Ser Met Ile Ile Gly Gly Ile Asp His Ser Leu Tyr
210 215 220

Thr Gly Ser Leu Trp Tyr Thr Pro Ile Arg Arg Glu Trp Tyr Tyr Glu
225 230 235 240

Sunesis-0021.ST25.txt

Val Ile Ile Val Arg Val Glu Ile Asn Gly Gln Asp Leu Lys Met Asp
245 250 255

Cys Lys Glu Tyr Asn Tyr Asp Lys Ser Ile Val Asp Ser Gly Thr Thr
260 265 270

Asn Leu Arg Leu Pro Lys Lys Val Phe Glu Ala Ala Val Lys Ser Ile
275 280 285

Lys Ala Ala Ser Ser Thr Glu Lys Phe Pro Asp Gly Phe Trp Leu Gly
290 295 300

Glu Gln Leu Val Cys Trp Gln Ala Gly Thr Thr Pro Trp Asn Ile Phe
305 310 315 320

Pro Val Ile Ser Leu Tyr Leu Met Gly Glu Val Thr Asn Gln Ser Phe
325 330 335

Arg Ile Thr Ile Leu Pro Gln Gln Tyr Leu Arg Pro Val Glu Asp Val
340 345 350

Ala Thr Ser Gln Asp Asp Cys Tyr Lys Phe Ala Ile Ser Gln Ser Ser
355 360 365

Thr Gly Thr Val Met Gly Ala Val Ile Met Glu Gly Phe Tyr Val Val
370 375 380

Phe Asp Arg Ala Arg Lys Arg Ile Gly Phe Ala Val Ser Ala Cys His
385 390 395 400

Val His Asp Glu Phe Arg Thr Ala Ala Val Glu Gly Pro Phe Val Thr
405 410 415

Leu Asp Met Glu Asp Cys Gly Tyr Asn Ile Pro Gln Thr Asp Glu Ser
420 425 430

Thr

<210> 82

<211> 42

<212> DNA

<213> Oligonucleotide to introduce ELNL site into soluble human proBACE1

<400> 82

ctcttcgtcg gtctccaggt tcagttccag acccagtgga gc

42

<210> 83

<211> 27

Sunesis-0021.ST25.txt

<212> DNA
<213> Oligonucleotide to introduce EINL site into soluble human proBACE1

<400> 83
ggctctccagg ttgatttcca gacccag

27

<210> 84
<211> 433
<212> PRT
<213> Soluble human proBACE1 containing an engineered EINL site starting at position 21

<400> 84

Thr Gln His Gly Ile Arg Leu Pro Leu Arg Ser Gly Leu Gly Gly Ala
1 5 10 15

Pro Leu Gly Leu Glu Ile Asn Leu Glu Thr Asp Glu Glu Pro Glu Glu
20 25 30

Pro Gly Arg Arg Gly Ser Phe Val Glu Met Val Asp Asn Leu Arg Gly
35 40 45

Lys Ser Gly Gln Gly Tyr Tyr Val Glu Met Thr Val Gly Ser Pro Pro
50 55 60

Gln Thr Leu Asn Ile Leu Val Asp Thr Gly Ser Ser Asn Phe Ala Val
65 70 75 80

Gly Ala Ala Pro His Pro Phe Leu His Arg Tyr Tyr Gln Arg Gln Leu
85 90 95

Ser Ser Thr Tyr Arg Asp Leu Arg Lys Gly Val Tyr Val Pro Tyr Thr
100 105 110

Gln Gly Lys Trp Glu Gly Glu Leu Gly Thr Asp Leu Val Ser Ile Pro
115 120 125

His Gly Pro Asn Val Thr Val Arg Ala Asn Ile Ala Ala Ile Thr Glu
130 135 140

Ser Asp Lys Phe Phe Ile Asn Gly Ser Asn Trp Glu Gly Ile Leu Gly
145 150 155 160

Leu Ala Tyr Ala Glu Ile Ala Arg Pro Asp Asp Ser Leu Glu Pro Phe
165 170 175

Phe Asp Ser Leu Val Lys Gln Thr His Val Pro Asn Leu Phe Ser Leu
180 185 190

Gln Leu Cys Gly Ala Gly Phe Pro Leu Asn Gln Ser Glu Val Leu Ala
 195 200 205

Ser Val Gly Gly Ser Met Ile Ile Gly Gly Ile Asp His Ser Leu Tyr
 210 215 220

Thr Gly Ser Leu Trp Tyr Thr Pro Ile Arg Arg Glu Trp Tyr Tyr Glu
 225 230 235 240

Val Ile Ile Val Arg Val Glu Ile Asn Gly Gln Asp Leu Lys Met Asp
 245 250 255

Cys Lys Glu Tyr Asn Tyr Asp Lys Ser Ile Val Asp Ser Gly Thr Thr
 260 265 270

Asn Leu Arg Leu Pro Lys Lys Val Phe Glu Ala Ala Val Lys Ser Ile
 275 280 285

Lys Ala Ala Ser Ser Thr Glu Lys Phe Pro Asp Gly Phe Trp Leu Gly
 290 295 300

Glu Gln Leu Val Cys Trp Gln Ala Gly Thr Thr Pro Trp Asn Ile Phe
 305 310 315 320

Pro Val Ile Ser Leu Tyr Leu Met Gly Glu Val Thr Asn Gln Ser Phe
 325 330 335

Arg Ile Thr Ile Leu Pro Gln Gln Tyr Leu Arg Pro Val Glu Asp Val
 340 345 350

Ala Thr Ser Gln Asp Asp Cys Tyr Lys Phe Ala Ile Ser Gln Ser Ser
 355 360 365

Thr Gly Thr Val Met Gly Ala Val Ile Met Glu Gly Phe Tyr Val Val
 370 375 380

Phe Asp Arg Ala Arg Lys Arg Ile Gly Phe Ala Val Ser Ala Cys His
 385 390 395 400

Val His Asp Glu Phe Arg Thr Ala Ala Val Glu Gly Pro Phe Val Thr
 405 410 415

Leu Asp Met Glu Asp Cys Gly Tyr Asn Ile Pro Gln Thr Asp Glu Ser
 420 425 430

Thr

Sunesis-0021.ST25.txt

<210>	85	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for L91C mutatio of human BACE1	
<400>	85	
	gcctgtatcc acacagatgt tgagcgt	27
<210>	86	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for D93C mutation of human BACE1	
<400>	86	
	actgctgcct gtacacacca ggatggt	27
<210>	87	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for T133C mutation of human BACE1	
<400>	87	
	ttcccacttg ccacaggtgt agggcac	27
<210>	88	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for Q134C mutation of human BACE1	
<400>	88	
	ttcccacttg ccacaggtgt agggcac	27
<210>	89	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for K168C mutation of human BACE1	
<400>	89	
	gttgatgaag aaacagtctg attcagt	27
<210>	90	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for F169C mutation of human BACE1	
<400>	90	
	gccgttgatg aaacacttgt ctgattc	27
<210>	91	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for I171C mutation of human BACE1	
<400>	91	
	gttggagccg ttacagaaga acttgtc	27

sunesis-0021.ST25.txt

<210> 92
 <211> 27
 <212> DNA
 <213> Oligonucleotide for W176C mutation of human BACE1
 <400> 92
 caggatgcct tcacagttgg agccggtt 27

<210> 93
 <211> 27
 <212> DNA
 <213> Oligonucleotide for I179C mutation of human BACE1
 <400> 93
 ggccagcccc agacagcctt cccagtt 27

<210> 94
 <211> 27
 <212> DNA
 <213> Oligonucleotide for I187C mutation of human BACE1
 <400> 94
 gtcaggcctg gcacactcag cataggc 27

<210> 95
 <211> 27
 <212> DNA
 <213> Oligonucleotide for R189C mutation of human BACE1
 <400> 95
 ggagtcgtca ggacaggcaa tctcagc 27

<210> 96
 <211> 27
 <212> DNA
 <213> Oligonucleotide for Y259C mutation of human BACE1
 <400> 96
 gatcacctca taacaccact cccgccg 27

<210> 97
 <211> 27
 <212> DNA
 <213> Oligonucleotide for K285C mutation of human BACE1
 <400> 97
 gtccacaatg ctacagtcac agttgta 27

<210> 98
 <211> 27
 <212> DNA
 <213> Oligonucleotide for I287C mutation of human BACE1
 <400> 98
 gccactgtcc acacagctct tgtcata 27

Sunesis-0021.ST25.txt

<210> 99
 <211> 27
 <212> DNA
 <213> Oligonucleotide for D289C mutation of human BACE1
 <400> 99
 ggtggtgccca ctacacacaa tgctctt 27

<210> 100
 <211> 27
 <212> DNA
 <213> Oligonucleotide for T292C mutation of human BACE1
 <400> 100
 acgaagggtg gtacagccac tgtccac 27

<210> 101
 <211> 27
 <212> DNA
 <213> Oligonucleotide for N294C mutation of human BACE1
 <400> 101
 gggcaaacga agacaggtgg tgccact 27

<210> 102
 <211> 27
 <212> DNA
 <213> Oligonucleotide for R296C mutation of human BACE1
 <400> 102
 ttcttgggc aaacaaaggt tggtggt 27

<210> 103
 <211> 27
 <212> DNA
 <213> Oligonucleotide for G325C mutation of human BACE1
 <400> 103
 caccagctgc tccactagcc agaaacc 27

<210> 104
 <211> 27
 <212> DNA
 <213> Oligonucleotide for R368C mutation of human BACE1
 <400> 104
 atcttcact ggacacaggt attgctg 27

<210> 105
 <211> 27
 <212> DNA
 <213> Oligonucleotide for K382C mutation of human BACE1
 <400> 105
 tgagatggca aaacagtaac agtcgctc 27

Sunesis-0021.ST25.txt

<210>	106	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for S386C mutation of human BACE1	
<400>	106	
	cgtaggatgac tgacagatgg caaactt	27
<210>	107	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for T390C mutation of human BACE1	
<400>	107	
	cataacagtg ccacaggatg actgtga	27
<210>	108	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for V393C mutation of human BACE1	
<400>	108	
	cagctcccat acaagtgccc gtggatg	27
<210>	109	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for inactivating D90N mutation of human BACE1	
<400>	109	
	actgctgcct gtgttcacca ggatgtt	27
<210>	110	
<211>	27	
<212>	DNA	
<213>	Oligonucleotide for inactivating D289N mutation of human BACE1	
<400>	110	
	ggtggtgcca ctgttcacaa tgctctt	27